A study is reported to determine whether bar soaps, without antibacterial additives, spread bacteria. The results reveal that bacteria are not transferred by this means from person to person, nor does the soap support bacterial growth.

# BACTERIOLOGICAL STUDIES RELATING TO HANDWASHING

### I. THE INABILITY OF SOAP BARS TO TRANSMIT BACTERIA

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FOLLOWING the first awareness of antibiotic resistant staphylococci in 1947, there has been an intensive epidemiologic study of the evolution, transmission, and importance of "hospital-acquired" diseases. As a result of these studies, the most often cited factors contributing to the "staph problem" usually include the emergence of antibiotic-resistant strains of Staphylococcus aureus following wide and sometimes indiscriminate clinical use of antibiotics, and the endemic nature of this organism in many hospitals.

It is generally concluded that there is no one way in which pathogenic or potentially pathogenic bacteria are disseminated through medical institutions. This observation has been borne out in the variety of areas that have been reported to harbor potentially pathogenic organisms. Since the successful control of hospital-related diseases depends on a renaissance of adequate hospital sanitation, as well as an awareness of bacterial foci, considerable attention has been given to locating objects and areas throughout the institutions that afford possible sources of

pathogenic or potentially pathogenic bacteria.

Many of the studies reporting the source of pathogenic bacteria have been based on the detection of nonspecific strains of Staph. aureus, and unfortunately have created undue concern regarding some inanimate objects by equivocally labeling them as "potential" or "possible" sources of pathogenic bacteria. Staph. aureus is shed readily by a large proportion of the populace<sup>7-16</sup> and can be found almost anywhere in the environment in contact with man. Considering the wide distribution of the organism, the incidence of staphylococcal infections is actually relatively infrequent.17,18

Although Staph. aureus as a group is considered pathogenic, antibiotic-resistant and classical pathogenic strains require further identification; i.e., phage typing, coagulase activity, antibiotic sensitivity spectrum, etc. 4,5,16,18,19 As a result of the detection of ubiquitous bacteria (i.e. Staphylococcus sp or Pseudomonas sp), pathogenic organisms have been reported to be found on bedding, floors, eating utensils, bathtubs, house-

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flies, air ducts, water carafes, mops, floor wax, sinks, water faucets, telephones, domestic animals, facial tissues (not used) and even bank notes. No clinical significance, however, has ever been established for most of the inanimate objects shown to be occasionally contaminated with Staph. aureus. For all practical purposes; the only reservoir for this organism is man.<sup>17</sup> The movement and dissemination of bacteria have been correlated with activities of human carriers (i.e., physicians, hospital personnel and hospital visitors) and with air currents associated with everyday hospital activity (i.e., bed making, mopping, etc.).

In connection with locating possible foci associated with nosocomial infections, it has been occasionally speculated that soap bars might possibly act as fomites in the dissemination of pathogenic bacteria. A search of the literature has failed to disclose any definitive report indicating that a bar of soap can spread bacteria after being used by an infected person or a carrier of an infectious bacteria. Conversely, Price<sup>20</sup> has found that neither unsterilized soap nor contaminated water affects the degree of skin degerming attained when using a bar soap (without antibacterial additives) in a surgical scrub procedure. Since Koch first reported the "bactericidal" properties of soap in 1881, there has been considerable work reported on the antibacterial nature of soaps. Although soaps in general show varying degrees of both bacteriostatic and bactericidal activity against a wide spectrum of microorganisms, under most practical-usage conditions, soap's chief advantage is its ability to suspend and wash away bacteria, as it does particulate soil. 10,21-24

In view of the inherent antibacterial nature of soap, as well as its ability to suspend and remove bacteria from the surface of the skin, it does not appear reasonable that a bar of soap would serve as a means for the transmission of bacteria. A series of studies has been performed to determine what role, if any, bar soap does play in the dissemination of microorganisms. A widely used pure soap bar without antimicrobial additives was employed.\* The study consisted of three parts as follows:

### Self-Sterilizing Capacity of Bar Soap (without antibacterial additives)

Several representative types of bacteria, including organisms isolated from the skin, were exposed to the surface of a bar of soap and sampled after varying lengths of time to determine the viability of the organism in contact with the surface of a bar soap (without antibacterial additives).

### Methods

A rectangular trough (approximately 2 cm x 4 cm x 2 mm deep) was cut into the surface of each of a series of small soap bars. These bars were then placed in a saturated humidity chamber (25° C) overnight to assure a uniform moisture content of the bar surface. After 18-24 hours, the conditioned bars were removed from the humidity chamber, and a 0.5 ml aliquot of washed cell suspension (24-hour culture) of the test organism was placed in each trough. The inoculated bars were then agitated to increase the contact between bacteria and soap surface and quickly replaced in the humidity chamber. After 2, 4, 6, 8, 10, 15 and 20-minute intervals, a bar was removed, and the trough contents were emptied into 100 ml sterile distilled water containing 0.5 per cent Tween 80. The addition of Tween 80 was found to retard curd formation and reduce foaming encountered during mixing (controls showed no appreciable antibacterial activity for Tween 80). The number of surviving bacteria was

<sup>\*</sup> Ivory.

calculated for each time increment using standard plate counts<sup>25</sup> on serial dilutions of this material.

### Results

The organisms employed in these experiments included: Staph. aureus (ATCC No. 6538), Escherichia coli (ATCC No. 10536) and two Gram-positive skin isolates (micrococci) having distinctly different biochemical characteristics. The relative abilities of these organisms to survive on the surface of a soap bar are shown by the data summarized in Figure 1. The results show a marked reduction of recoverable viable organisms within a few minutes' ex-

posure to soap. In spite of considerable variation in effectiveness, all the bacterial species were reduced in number by at least five logarithm units (100,000-fold) after 15-minute exposure. These data reaffirm the bactericidal nature of soap per se. Obviously, none of the test organisms will increase in number on the surface of the bar soap.

## 2. Transmissibility of a Tracer Bacterium by Bar Soap

In this series of experiments the use of a tracer organism was employed to determine (a) the extent of transfer of bacteria from very heavily contami-

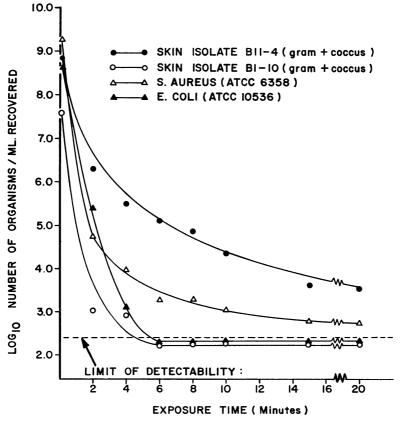


Figure 1—The Viability of Four Organisms when Exposed to the Surface of Bar Soap

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nated skin to the surface of a bar of soap during washing of the hands, and (b) whether immediate subsequent use of the bar by a second person results in the transfer of these organisms to his Serratia marcescens, Gram-negative, constant pigmenting mutant not ordinarily found on the skin, was chosen as the tracer organism because (1) it can be enumerated easily and differentiated (by a bright red pigment) from the organisms normally encountered on the skin, (2) it adheres readily to the skin, and (3) it is less affected by soap than either Staph. aureus or E. coli as determined by comparative bacteriostatic breakpoint (minimal inhibitory concentration) tests.

### Method A

The first series of tests was designed to determine whether organisms could be transferred (under experimental conditions) from the skin to the surface of a soap bar during handwashing. Each panelist immersed one hand for one minute in a dilution of a 24-hour culture of Serratia, containing approximately 2 x 109 organisms/ml. It was estimated that approximately 5 x 109 organisms (Serratia) remained on the skin after this contamination. The panelist then washed his hands with soap, using his normal washing procedure. The used soap bar and the previously contaminated hand were both sampled quantitatively for the presence of the tracer organism at the end of the handwashing operation.

The used bar at the completion of the handwashing was placed directly in 100 ml of sterile distilled water where it was agitated for five minutes. This time of agitation had been previously shown to be sufficient to remove quantitatively the tracer organism from the bar. The total number of bacteria on the surface of the used soap bar was calculated from a standard plate count of the 100 ml of water.

Table 1—Distribution of a Tracer Organism (Serratia marcescens) after Washing\* a Heavily Contaminated Hand with Bar Soap

	Total Number of Serratia Recovered		
Panelist	Washed Hand†	Soap Bar After Washing	
1	$3.2 \times 10^5$	$2.0 \times 10^3$	
2	$8.6 \times 10^{5}$	$3.5 \times 10^{4}$	
3	$2.8 \times 10^{5}$	$3.0 \times 10^5$	
4	$6.0 \times 10^{4}$	$2.0 \times 10^{5}$	
5	$2.0\times10^{6}$	$9.0 \times 10^5$	
6	$1.4 \times 10^5$	$2.3 \times 10^5$	
7	$2.8 \times 10^6$	$3.0 \times 10^{6}$	
8	$1.1 \times 10^{6}$		
9	$1.0 \times 10^{7}$	$1.3 \times 10^{4}$	
10	$1.7 \times 10^4$	$9.0 \times 10^{5}$	
Mean %			
Reduction	99+%‡	$6.2 \times 10^5$	

<sup>\*</sup>Subject washed hands in his customary manner. † Previous to washing, one hand was contaminated by a one-minute dip in a dilution of a 24-hour culture of Serratia marcescens var. 8-uk containing approximately 2.0×10° organisms/ml.

t Mean per cent reduction of Serratia from the hands based on an approximation of the original contamination level and the level recovered after washing.

The number of bacteria remaining on the contaminated hand after it had been washed was determined as follows: A sterile surgical glove was placed on the previously contaminated hand and 20 ml of sterile trypticase soy broth was poured into the glove, where it was allowed to remain in contact with the hand for five minutes. During this five-minute period, the hand was "worked" in the glove to increase contact and agitation between the hand and the culture medium. The broth was then removed, and the number of Serratia marcescens organisms was determined by standard plate count (surface streak method). Data from these tests are presented in Table 1.

### Method B

A second series of experiments to determine the transferability of the tracer organism from person to person via bar soap followed the same general procedure described above with the following modifications. The soap bar was used for handwashing by the first panelist, both of whose hands had been heavily contaminated with the tracer bacteria (as in Method A). It was then picked up and used by a second panelist within one minute of the first use. The number of bacteria remaining on the bar after use by the second panelist and those redeposited on his hands were determined by standard plate counting methods, as described above.

The data from these tests are presented in Table 2.

#### Results

The data presented in Table 1 show that some organisms can be transferred to the surface of a bar of soap during washing of a massively contaminated skin area.

The data sumarized in Table 2 show, however, that there is no transfer of organisms to the skin of a second person who washes with the bar immediately after use by the massively contaminated first user. In addition to demonstrating the inability of soap to transfer this organism from one person to another, the tests also show that most of the bacteria previously deposited on the soap bar are eliminated during one subsequent use of the bar for handwashing.

### 3. Accumulation of Bacteria on Soap During Actual Usage

A series of studies was designed to determine whether any appreciable deposition or build-up of bacteria on the surface of a soap bar occurred during actual heavy use.

### Methods

The level of viable bacteria on the surface of the soap after intensive use in the washrooms of a large office-laboratory building during a seven-hour period was determined. Soap bars were placed in the washrooms as ordinarily

dispensed; no attempt was made to protect them from atmosphere, dust, etc. At the end of the seven-hour usage period, the bars were collected in sterile petri dishes. Three discs (1.7 cm diameter x approximately 1.5 mm deep) were aseptically removed from two sides of each bar soap. The six discs from each bar were combined and dissolved in 50 ml of sterile distilled water. Standard trypticase soy agar plate counts were made from this solution.

The level of contamination on the surface of the soap was expressed as the number of bacteria per square centimeter of soap surface. The average number of times the bar was used was estimated from a count of the number of hand towels used during the test period.

Table 2—The Inability of Bar Soap to Transmit a Tracer Bacterium (Serratia marcescens) from Heavily Contaminated Hands to the Hands of Another User

(Approximate Bacterial Level on Bar Used by Second Panelist=6×10<sup>5</sup> Organisms)

Soap Bar	S. marcescens Transferred to Second Panelist* (Total No. Recovered) for Both Hands. Limit of Detectability 20 Organisms.	Residual on Soap Bar after Use by Second Panelist (Total No. Recovered). Limit of Detectability 100 Organisms.
1	0	3.8×10 <sup>4</sup> †
2	0	0
3	0	0
4	0	0
5	0	0
6	0	0
7	0	0
8	0	0
9	0	0
10	0	0

\*Just prior to use by the second panelist, the bar was contaminated (See Method B, page 918).
†This high recovery of S. marcescens probably resulted from unusually high contamination of an area on the soap bar (i.e., the end) not diluted during use by second panelist.

Table 3—The Level of Viable Bacteria Isolated from the Surface of Bar Soap after Actual Use\*

(The soap bars were placed in drainable or nondrainable receptacles between use)

Soap Bar	Bar Soap Receptacle	Bacterial Accumulatio (Organisms, Sq Cm)	
1	Drainable Ty	pe 27	28
	,, ,,	0†	28
$\frac{2}{3}$	" "	0	28
	" "	0	28
4 5	" "	0	84
6	, ,, ,,	0	84
7	" "	0	84
8	" "	0	84
9	Nondrainable Ty	pe 0	84
10	"	7	84
11	" "	44	84
12	" "	104	84
13	" "	0	105
14	"	68	105
15	"	0	105
16	"	0	105

<sup>\*</sup>The level of bacterial contamination following actual usage was determined on soap and soap receptacles from washrooms of a large office building.
†Limit of detectibility four organisms/Sq Cm.

### Results

The frequency of usage of the bars was such that little or no drying occurred, and a reduction in viable organisms as a result of dehydration was probably much less than that encountered in moderate usage. The data in Table 3 indicate that, although bar soap kept in use in nondrainable-type soap dishes has a tendency to accumulate more bacteria than bars kept in receptacles that drain, it is evident that with either type dish an appreciable build-up of microorganisms on soap bars does not occur as a result of repeated handwashing.

### Discussion

Historically, soap has established its usefulness as a major contributor to modern hygiene and hospital sanitation. Within recent years, a number of bar and liquid skin-cleaning soap and detergent products containing bacteriostatic additives designed to extend the already existing antibacterial properties of soap have been introduced. In the use of these sanitizer-containing products, however, it should not be overlooked that probably the most important single factor in the control of undesirable microorganism is cleanliness. Antibacterial additives can supplement but cannot replace the mechanical removal of bacteria and dirt resulting from good cleaning practice.<sup>22,23</sup>

In view of the lack of experimental information, it is conceivable that misconceptions regarding the role of soap itself and of the bar form in the prevention of disease could develop. It is with this possibility in mind that the authors have undertaken to evaluate

critically the role of soap bars in the dissemination of bacteria.

The results of our tests with a tracer organism show that bacteria are not transmitted from person to person through the use of a commercial bar of soap. The hands of the panelists artificially contaminated with approximately 5 x 109 organisms, which represent about 1,000 times the number of bacteria usually recovered from hands during a single thorough handwashing, did not result in the transfer of the tracer organism to the hands of a second person using the same soap bar.

These observations verify some of the logical arguments long used to describe the function of bar soap as a sanitizing agent: (1) that bacteria are physically eliminated from the skin during regular usage of soap in much the same manner as particulate soil and (2) that bacteria are not redeposited on the skin of subsequent users any more than particulate dirt would be redeposited.

The surface of regular bar soaps unordinary usage conditions can hardly be expected to contain the necessary nutrients and growth factors required for the proliferation of most pathogenic organisms, particularly Staph. aureus. In addition, many adverse physical factors also lessen the likelihood of bacteria surviving on the surface of bar soaps (i.e., desiccation, temperature, etc.). The results of the tests, reported here on the level of bacteria recovered from the surface of soap bars after actual usage or artificial contamination, demonstrate that bar soap is inherently antibacterial and show that it would not likely support the growth of bacteria.

In view of the demonstrated inability of bar soap to transfer bacteria to the skin of users from unrealistically high levels of tracer organisms deposited on its surface, the relatively low level of bacterial contamination sometimes found on bar soaps in use would appear to pose no threat to subsequent users. The level of nonpathogenic bacteria found on the surface of bar soap under heavy conditions is not surprising (Table 3), since it is not uncommon to remove more than 2 x 106 organisms from the skin in a single handwashing.

### Conclusions and Summary

Several bacteriological tests designed to evaluate the role of bar soaps (without antibacterial additives) in the spread of bacteria have demonstrated the following:

- 1. Bacteria are not transferred from person to person through the use of bar soaps.
- 2. Bar soaps do not support the growth of bacteria under usage conditions.
- 3. Bar soaps are inherently antibacterial by their physical-chemical nature.
- 4. The level of bacteria that may occur on bar soap even under extreme usage conditions (heavy usage, poorly designed nondrainable soap dishes, etc.) does not constitute a health hazard.

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This is the first of a series of studies to determine what role, if any, bar soap plays in the dissemination of microorganisms. The paper was submitted for publication in December, 1963.

### To Alicia:

What of the future world, what will it bring?

More people than the world can then support.

This is the raw refrain of each report

The everlasting statisticians sing.

No place to stand, no place to sit, no thing
to eat or drink, no fun of any sort;

No place to park, nor any room for sport

In woods or fields, come summer, fall or spring.

Then if we cannot save the world from this

Must we like lemmings scurry down to sea

Accepting with a laugh our watery fate?

If so, then grant me dear one final bliss

And say that you will scurry down with me.

But tell me now before it is too late.

### Homer N. Calver

(P.S. I do not know any Alicia. Sonnets always seem to be to some lady or other and Alicia sounds romantic. Also begins with "A" to get listed first in the index!)